EJN fens EUROPEAN JOURNAL OF NEUROSCIENCE

European Journal of Neuroscience, Vol. 33, pp. 371–381, 2011 doi:10.1111/j.1460-9568.2010.07504.x

BEHAVIORAL NEUROSCIENCE

Mammillothalamic tract lesions disrupt dead reckoning in the rat

Shawn S. Winter, Steven J. Wagner, Jeffery L. McMillin and Douglas G. Wallace
Psychology Department, Northern Illinois University, DeKalb, IL 60115-2892, USA

Keywords: limbic system, mammillary bodies, path integration, spatial orientation, translational neuroscience

Abstract

Debate surrounds the role of the limbic system structures' contribution to spatial orientation. The results from previous studies have supported a role for the mammillary bodies and their projections to the anterior thalamus in rapid encoding of relationships among environmental cues; however, this work is based on behavioral tasks in which environmental and self-movement cues could not be dissociated. The present study examines the effects of mammillothalamic tract lesions on spatial orientation in the food hoarding paradigm and the water maze. Although the food hoarding paradigm dissociates the use of environmental and self-movement cues, both sources of information are available to guide performance in the water maze. Mammillothalamic tract lesions selectively impaired performance on both tasks. These impairments are interpreted as providing further evidence for the role of limbic system structures in processing self-movement cues.

Introduction

Spatial orientation is the ability to guide movements through an environment, and requires processing of multiple sources of information (Gallistel, 1990). The source of information can be classified based on whether it originates from the environment or is generated as a result of the animal's movement. As an animal becomes familiar with an environment, it may learn to directly follow environmental cues (i.e., beacon homing) or learn to move relative to environmental cues (i.e., piloting; Means *et al.*, 1992; Pearce *et al.*, 1998; Wallace *et al.*, 2002a). Recent work has demonstrated that within a single movement trajectory, both humans and rats sequentially engage in piloting and then beacon homing strategies (Hamilton *et al.*, 2004, 2009). Animals also generate cues (i.e., vestibular, proprioceptive, optic flow, or motor efferent copies) as a result of moving through an environment. If access to familiar environmental cues is limited (e.g., at night) or the environment is novel, an animal can use self-movement cues to update an online representation of its current position. Use of this online representation to estimate direction and distance to the location from where movement originated is referred to as dead reckoning (Darwin, 1873; Murphy, 1873; Barlow, 1964; Etienne & Jeffery, 2004; Wallace *et al.*, 2008). Continuous processing of environmental and self-movement cues ensures that an animal can maintain spatial orientation under varying environmental conditions.

Convergence of environmental and self-movement cue processing has been posited to occur within the limbic system. Both sources of information have been shown to influence the firing characteristics of head direction cells in the limbic system. Salient environmental cues have been shown to control directional tuning of head direction cells (Taube *et al.*, 1990b; Blair & Sharp, 1996). In addition, self-movement cues have been shown to be sufficient to maintain the head direction cell signal under dark conditions (Goodridge *et al.*, 1998) and in the presence of rapidly rotating environmental cues (Blair & Sharp, 1996). Generation of the head direction cell signal depends in part on self-movement cues originating from stimulation of the vestibular system (Stackman & Taube, 1997) and subsequent processing by the dorsal tegmental nucleus of Gudden (DTN) and the lateral mammillary nucleus (Bassett *et al.*, 2007). Directional tuning of head direction cells by environmental cues is mediated by reciprocal connections of the anterodorsal thalamic nucleus (ADN) with the postsubiculum (Goodridge & Taube, 1997) and the retrosplenial cortex (Clark *et al.*, 2010). Observing that each source of information has effects on head direction cell firing characteristics that are location-dependant is consistent with work demonstrating that lesions of the DTN and ADN differentially influence spatial orientation (Frohardt *et al.*, 2006). Specifically, DTN lesions severely disrupted performance on spatial tasks in which rats had access to both sources of information or were restricted to self-movement cues, whereas ADN lesions only produced mild impairments on either spatial task. The mammillary bodies (MB) have reciprocal connections with the DTN and send projections to the ADN; therefore, the MB are likely to be critically involved in maintaining spatial orientation.

Previous work has demonstrated that MB lesions disrupt performance on spatial tasks (Sutherland & Rodriguez, 1989; Neave *et al.*, 1997; Vann & Aggleton, 2003; Vann, 2005); however, two factors have limited the ability to characterize the role of the MB in maintaining spatial orientation. First, the supramammillary nucleus (SUM) is immediately dorsal to the MB and has been shown to be involved in the generation of hippocampal theta (Kirk *et al.*, 1996;

Correspondence: D. G. Wallace, as above.
E-mail: dwallace@niu.edu

Received 23 July 2010, revised 6 October 2010, accepted 11 October 2010

© 2010 The Authors. European Journal of Neuroscience © 2010 Federation of European Neuroscience Societies and Blackwell Publishing Ltd

Kirk, 1997). Therefore, impaired performance on spatial tasks subsequent to MB lesions may depend on damage to surrounding structures, including the SUM. In contrast, transection of the mammillothalamic tract (MTT) eliminates MB projections to the ADN while sparing the SUM, providing a more selective technique for investigating the role of the MB in spatial orientation. Second, although the effects of more selective lesions have been attributed to impaired mnemonic function (Vann & Aggleton, 2003; Vann, 2005), the behavioral tasks that have been used fail to dissociate the use of different navigational strategies. Therefore, performance associated with more selective lesions may reflect impaired mnemonic function, impaired processing of environmental or self-movement cues, or a combination of impairments. In light of these limitations, the present study uses the food hoarding paradigm (Whishaw & Tomie, 1997; Maaswinkel *et al.*, 1999; Wallace *et al.*, 2002b; Martin & Wallace, 2007) to characterize deficits in spatial orientation subsequent to MTT transection.

The food hoarding paradigm exploits the rat's natural tendency to carry food to a refuge for consumption to dissociate the use of different navigational strategies. First, in an illuminated environment with a cued refuge, rats may use proximal cues associated with the refuge (beacon homing), distal room cues (piloting) or self-movement cues (dead reckoning) to return to the refuge to consume the food item. Next, hiding the refuge under the edge of the table limits rats to using strategies based on piloting or dead reckoning to return to the refuge. Further, testing under completely dark conditions limits rat to using dead reckoning-based navigation to return to the refuge. Finally, moving the location of the hidden refuge around the edge of the table in an illuminated environment places piloting and dead reckoning strategies in conflict regarding the location of the refuge. Performance in the food hoarding paradigm characterizes the nature of the information processing deficits associated with MTT transection. Performance can also be assessed in the place training and matching-to-place tasks in the water maze (Whishaw, 1985) to provide a general assessment of spatial orientation.

Materials and methods

Animals

Thirty-six female Long Evans rats (*Rattus norvegicus*) obtained from Northern Illinois University vivarium were pair-housed in plastic cages. The colony room was maintained at ∼20–21 °C and on a 12-h light–dark cycle. Rats weighed ∼250 g at the beginning of the experiment. Throughout testing, rats were fed supplemental rat chow (2L42 Rodent Breeder Diet food Pellets; PMI Nutrition International, Brentwood, MO, USA) to maintain them at 85% of their free-feeding weight. In addition to daily feeding, rats were given several 1-g banana pellets before testing started to habituate the rats to the food items used in the experiment. The NIU Institutional Animal Care and Use Committee, which follows the guidelines set forth by the Office of Laboratory Animal Welfare, approved all procedures used in this experiment.

Surgery

All rats were deeply anesthetized with a mixture of isoflurane and oxygen during the surgery. Electrolytic lesions of the MTT were produced by passing a cathodal current of ∼1.0 mA for 5 s through an electrode insulated except for the tip. There was a single lesion site per hemisphere; coordinates with respect to bregma and the surface of the dura were: AP, −1.9 mm; ML, ±0.9 mm; DV, −6.6 mm. Pilot work demonstrated the difficulty in consistently producing bilateral MTT lesions; therefore, 27 rats were assigned to receive MTT lesions. Sham-operated rats (n = 9) were treated the same except the electrode was lowered 5.6 mm below the surface of the dura without passing the current through the electrode. Food hoarding began ∼4 weeks after the surgery.

Apparatus

Food hoarding table

The apparatus was a wooden circular (200-cm-diameter) table that was painted white (see Fig. 1A). The surface of the table was ∼100 cm above the floor. The cued refuge was a Plexiglas box

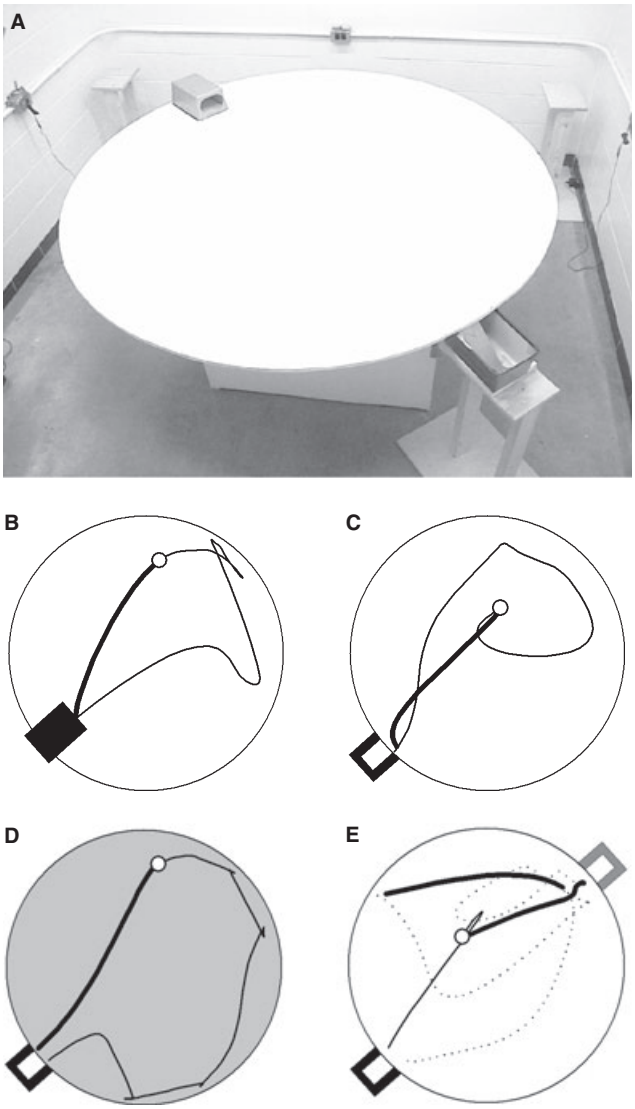

FIG. 1. (A) Photograph of the testing apparatus presented under light conditions with the cued refuge and hidden refuge located at the periphery of the table. Representative food hoarding trips are presented for (B) the cued training, (C) hidden probe, (D) dark probe and (E) reversal probe. The outward segments (thin line) are circuitous paths that end at the food pellet (white dot). The homeward segments (thick line) are non-circuitous paths directed towards the refuge. During the reversal probe, the rat makes several attempts to carry the food item to the former refuge location (grey square) prior to returning to the current refuge location (black square).

© 2010 The Authors. European Journal of Neuroscience © 2010 Federation of European Neuroscience Societies and Blackwell Publishing Ltd

(14 × 10 × 24 cm) with an 8 × 6 cm opening on the shorter end and was covered with duct tape to make it opaque (positioned in the northwestern quadrant of the table; Fig. 1). During probes, a second Plexiglas box of the same dimensions without the hole cut in one end was used as the refuge. The probe refuge had a short ramp leading onto the table and was supported under the surface of the table by a stand (positioned in the southeastern quadrant of the table; Fig. 1). The table was located in a square room that was prepared so that light would not penetrate when testing under dark conditions. To observe rat behavior under dark conditions, experimenters wore night-vision goggles, a night-vision camera was attached to the ceiling of the testing room, and infrared emitters were positioned along the walls. Previous studies have demonstrated that rats are not able to detect wavelengths within the infrared range (Neitz & Jacobs, 1986). The night-vision camera was connected to a DVD recorder located in an adjacent room providing a record of the rats' behavior for subsequent analysis.

Water maze

The water maze was an aluminum circular (173 cm diameter) tub with 58-cm-high walls. The circular escape platform (15 cm diameter) could be positioned in different locations in the maze. The maze was filled such that the water level was 2 cm above the surface of the escape platform. The water was maintained at 19 °C, and white non-toxic paint was added to make the water opaque. A ceiling-mounted camera was connected to a DVD recorder providing a record of the rats' behavior for subsequent analysis.

Procedure

Food hoarding

An opaque cage with a wire mesh top was used to transport rats from the colony to the testing room. During transportation, lights were turned off, the cage was rotated, and the experimenter walked a circuitous path that varied across days. This limited the rat's ability to learn the location of the testing room relative to the colony. During a training session (30 min), a rat was placed in the cued refuge and shaped to leave the refuge to search for a randomly located 1.0-g banana-flavored food pellet (BioServ, Frenchtown, NJ, USA) then carry it to the refuge. Daily training continued until a rat retrieved five randomly located food pellets within a session for five sessions. The last training session was recorded. Performance observed with the cued refuge is an index of the rat's ability to use proximal environmental cues (i.e., the refuge), distal environmental cues (i.e., landmarks in the room) or self-movement cues (i.e., vestibular, proprioception, optic flow, or efferent copies) to guide navigation (see Fig. 1B). After training sessions, rats experienced a sequence of probe sessions alternating with cued refuge sessions.

During cued refuge and probe sessions, each rat searched for a randomly located food pellet and carried it to the refuge prior to placement of the next food pellet. This continued until five food pellets were retrieved from the table. The hidden probe involved positioning the refuge below the surface of the table such that the rat could climb onto the table, yet the proximal cues associated with the refuge were minimized. The refuge remained in the same position in the room as experienced during training with the cued refuge. Performance on this probe is an index of the rat's ability to use distal environmental cues or self-movement cues to return to the refuge (see Fig. 1C). During the dark probe, the refuge was located just below the surface of the table and in the same position in the room as experienced during training with the cued probe. In addition, the testing room lights were turned off, thereby eliminating visual cues associated with the refuge. Performance on the dark probe is an index of the rat's ability to use self-movement cues to return to the refuge (see Fig. 1D). The reversal probe was identical to the hidden probe with the exception that the refuge was rotated 180° around the edge of the table, relative to the position experienced during training with the cued refuge. This probe places distal environmental and self-movement cues in conflict regarding the position of the refuge after locating the food pellet. If distal environmental cues are used to guide navigation then the rat should return to the former refuge location experienced during training; however, if self-movement cues are used to guide navigation the rat will return to the new refuge location (see Fig. 1E). After a rat completed its daily session, the table was wiped down with ammonia-based cleaner and the table was rotated varying degrees; both actions were intended to minimize the ability of rats to learn to use odor cues to guide performance (Wallace *et al.*, 2002a).

Water maze

Opaque cages with wire mesh tops were used to transport rats from the colony to the testing room. During a place training trial, a rat was placed in the water maze at one of the cardinal compass directions facing the apparatus wall. The rat swam until locating the hidden platform or 60 s had elapsed. If the rat located the platform before 60 s then it remained on the platform for 30 s. If the rat failed to locate the platform before 60 s then it was guided to the platform where it remained for 30 s. After the rat was removed from the platform, it was dried with a towel and returned to the transport cage. This procedure continued until all rats were released from all four cardinal compass directions. Considering that rats were run in squads of ∼10, intertrial intervals were initially 30 min and then gradually decreased as the performance improved across the five training days. Matching-to-place testing started the day after the last place training trial and lasted 4 days. During matching-to-place testing, the hidden platform shifted to the center of a new quadrant each day while the position remained stable within a day. The trial procedures were the same as described above with the exception that rats only received two trials per day. The water was stirred between trials and replaced after each day to limit the rats' ability to use odor cues to guide performance (Means *et al.*, 1992).

Data analysis

The Peak Perfomance (Vicon, Denver, CO, USA) motion capture system was used to quantify movement characteristics of rats in the food hoarding paradigm. Rat movement was tracked by selecting one pixel every fifth frame that corresponded to the midline of the body at the level of the forelimbs. The resulting *x*- and *y*-coordinates were scaled to real-world units and used to calculate the rats' moment-to-moment speeds. Each trial was divided into outward and homeward segments. The outward segment was defined as all of the movements that displaced the rat from the refuge until locating the food pellet. Two measures were used to characterize the outward segment: path circuity and time elapsed. Path circuity was calculated by dividing the distance between the start and end points of the path by the total distance traveled. Time elapsed was calculated as the duration of the outward segment. Mixed-design ANOVAs with Lesion (between-subjects) and Trip (within-subject) as factors were conducted on both measures.

The homeward segment was defined as all of the movements occurring after the rat located the food pellet until returning to the

© 2010 The Authors. European Journal of Neuroscience © 2010 Federation of European Neuroscience Societies and Blackwell Publishing Ltd

refuge. Two measures were used to characterize performance on the homeward segment: path circuity and heading direction. Path circuity on the homeward segment was calculated identically to the outward segment. A mixed-design ANOVA with Lesion (between-subjects) and Trip (within-subject) as factors was conducted on path circuity. Heading direction was calculated as the angle subtended by the following points: center of the refuge, food pellet location, and midpoint of the homeward segment. Heading directions have a circular distribution, ranging from 0° (centered on refuge) increasing counterclockwise to 359° (1° left of the refuge). Linear statistics fail to capture the quantitative similarity of 0° and 359°; descriptive and inferential circular statistics techniques have been developed to characterize data with a circular distribution (Batschelet, 1981). Heading directions are first evaluated for group differences in variability or clustering. The length of the mean vector is a descriptive statistic that characterizes the clustering associated with a set of heading directions ($r = 0.0$, headings are randomly distributed; $r = 1.0$, headings are the same value). If groups are significantly different in the average length of their mean vector, further analysis of group differences based on average heading direction is precluded. Provided that the average length of the mean vector does not significantly differ between groups, then the Watson–Williams F-test can be applied to evaluate whether groups differ based on average heading direction.

The EthoVision (Noldus, Leesburg, VA, USA) motion capture system was used to quantify movement characteristics of rats in the water maze. Latency to reach the platform was calculated for each trial. During place training, blocks reflected the average latency to locate the hidden platform observed across four trials. During matching-to-place testing, the latency to locate the hidden platform on the first (block 1) and second (block 2) trials was averaged across days. Mixed-design ANOVAs with Lesion (between-subjects) and Block (within-subject) as factors were conducted on place training and matching-to-place latency data.

Histology

Upon completion of behavioral testing all rats were perfused through the heart, first with phosphate-buffered saline, then 4.0% paraformaldahyde. The brain was extracted and soaked for 24 h in 4.0% paraformaldahyde, followed by 24 h in 30% sucrose solution. Next, the brains were frozen and sliced using a cryostat (Global Medical Instruments, Inc., Ramsey, MN, USA). Two sets of coronal sections were taken for histological analysis. One set was sliced at 50 μm and processed for Cresyl violet stain to assess transection of the MTT. Previous work has demonstrated that MB damage significantly alters hippocampal cholinergic function (Béracochéa *et al.*, 1995). The other set was sliced at 40 μm and processed for acetylcholinesterase (AChE) to assess the effects of MTT transection on hippocampal cholinergic function. Digital photographs were taken of coronal sections at the level of the dorsal hippocampus (~ -2.7 mm relative to bregma). These files were converted to grey-scale and opened with Scion Image for Windows (Scion Corporation, USA; freely available on the internet at http://www.scioncorp.com). Martin & Wallace (2007) used optical density of AChE stained sections to characterize the cholinergic function of the hippocampus (including CA1, CA3 and DG regions) and overlying cortex (including somatosensory and motor cortex) subsequent to 192 IgG-Saporin lesions of the medial septum. The current study obtained optical density values (white, 0.0; black, 255) from rectangular areas (50×60 pixels) within the same brain regions.

Results

Histology

Photomicrographs are presented for representative rats receiving Sham (see panels A and B of Fig. 2) or Bilateral (see panels C–H) lesions. Histological analysis of lesion extent revealed variability in the bilateral destruction of the MTT. Damage to the MTT was characterized as either complete (i.e., no visual evidence of tract encapsulation, collapsing of surrounding tissue, and presence of gliosis) or some level of sparing for each hemisphere. Application of these criteria resulted in three groups: the Miss group had some level of bilateral sparing of the MTT ($n = 10$; see panels C and D of Fig. 2); the Unilateral group had some level of sparing of the MTT restricted to one hemisphere ($n = 12$; see panels E and F of Fig. 2); and the Bilateral group did not have any sparing of the MTT bilaterally ($n = 5$; see panels G and H of Fig. 2). No significant differences were observed between the Sham and Miss groups on food hoarding or water maze measures; therefore, the two groups were combined into a single group. All subsequent analyses were conducted with the following groups: Sham/Miss ($n = 19$), Unilateral ($n = 12$) and Bilateral ($n = 5$).

Previous research has shown that MB lesions alter hippocampal cholinergic function (Béracochéa *et al.*, 1995); however, MTT transection spares MB and adjacent structures (i.e., SUM) that influence hippocampal function. These factors prompted assessment of hippocampal and cortical cholinergic function. AChE-stained brain sections were only available for a subset of rats; therefore, quantification of AChE optical density was limited to: Sham/Miss ($n = 16$), Unilateral ($n = 8$) and Bilateral ($n = 5$) rats. A general lightening of the AChE stain was evident in the rats with bilateral MTT lesions (see Fig. 3). The ANOVA conducted on AChE optical densities in the hippocampus revealed a significant effect of Group ($F_{2,28} = 6.1$, $P = 0.006$). *Post hoc* analysis revealed that the Bilateral group had significantly lower optical density scores than did the Sham/Miss and Unilateral groups ($P < 0.05$). The ANOVA conducted on optical densities in the cortex failed to reveal a significant effect of Group ($F_{2,28} = 2.2$, $P = 0.130$).

Food hoarding

Cued refuge

When provided with the cued refuge, all rats carried the food pellet to the refuge for consumption. The topographic and temporal characteristics of the outward segment did not vary among groups. The ANOVA conducted on path circuity failed to reveal a significant effect of Group ($F_{2,33} = 1.7$, $P = 0.198$), Trip ($F_{4,132} = 1.1$, $P = 0.359$) or Group × Trip interaction ($F_{8,132} = 0.9$, $P = 0.466$). The ANOVA conducted on time elapsed failed to reveal a significant effect of Group ($F_{2,33} = 0.7$, $P = 0.504$), Trip ($F_{4,132} = 0.8$, $P = 0.527$), or Group × Trip interaction ($F_{8,132} = 1.2$, $P = 0.304$). Groups did not significantly differ in the organization of their outward segments.

Topographic characteristics of the homeward segment did not vary among groups (see panel A of Fig. 4). The ANOVA conducted on path circuity failed to reveal a significant effect of Group ($F_{2,33} = 2.5$, $P = 0.098$), Trip ($F_{4,132} = 1.1$, $P = 0.359$), or Group × Trip interaction ($F_{8,132} = 1.4$, $P = 0.202$).

Circular statistics were used to characterize group differences in heading direction associated with the cued refuge. Mean vector length (extent that heading directions are clustered around one direction) and average heading direction were calculated for a rat's set of heading directions. The ANOVA conducted on mean vector length failed to reveal a significant effect of Group ($F_{2,33} = 0.7$, $P = 0.504$). This

© 2010 The Authors. European Journal of Neuroscience © 2010 Federation of European Neuroscience Societies and Blackwell Publishing Ltd

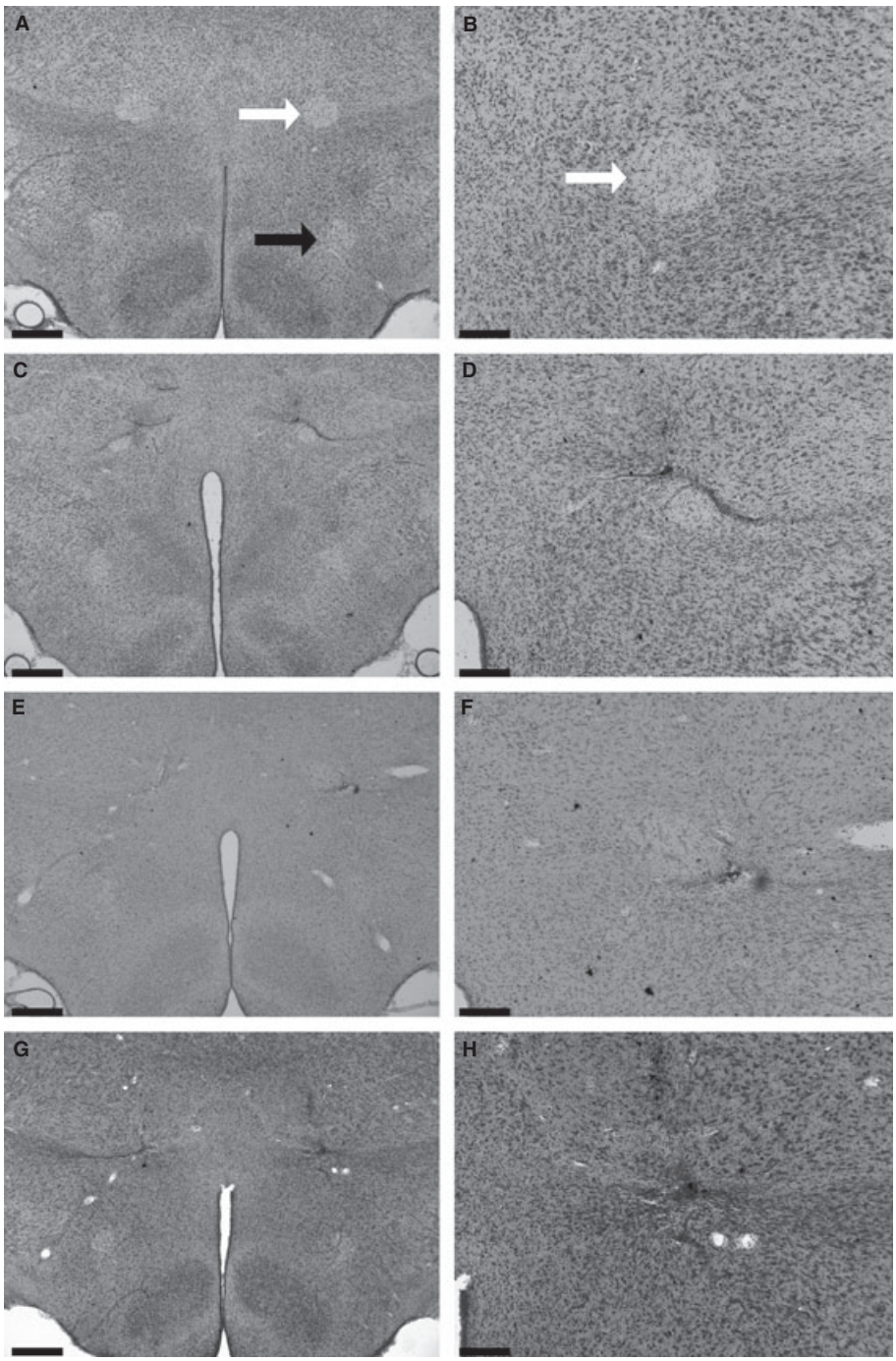

FIG. 2. Cresyl violet-stained coronal sections are provided for representative rats from (A and B) the Sham, (C and D) Miss, (E and F) Unilateral and (G and H) Bilateral groups. The left-hand panels are at lower magnification and characterize the mammillothalamic tract (white arrow) and the fornix (black arrow) at the level of maximum lesion extent. The right-hand panels are of the same sections, taken at higher magnification, and focus on the damage sustained to the mammillothlamic tract. Any sparing of the mammillothalamic tract bilaterally was classified as a Miss (panels C and D), whereas any sparing of the mammillothalimic tract in one hemisphere was classified as Unilateral (panels E and F). Scale bars, 500 μm (left-hand panels), 200 μm (right-hand panels).

result permitted the use of circular statistics to assess group differences in average heading direction. The Watson–Williams F-test revealed a significant effect of Group ($F_{2,33} = 4.1$, $P = 0.026$). Subsequent analyses revealed that the Sham/Miss group's average heading direction significantly differed from that of the Unilateral ($F_{1,29} = 6.8$, $P = 0.014$) and Bilateral ($F_{1,22} = 6.5$, $P = 0.018$) groups. Although significant differences were observed in the average heading direction among groups, all groups were orientated towards the refuge (see panel A of Fig. 5).

Hidden probe

During the hidden probe, all rats carried the food pellet to the refuge for consumption. The topographic and temporal characteristics of the outward segment did not vary among groups. The ANOVA conducted

© 2010 The Authors. European Journal of Neuroscience © 2010 Federation of European Neuroscience Societies and Blackwell Publishing Ltd

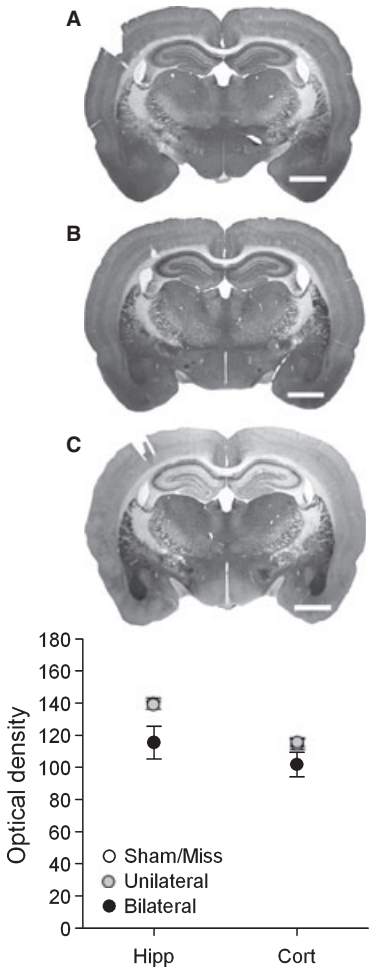

FIG. 3. Sections stained for AChE are presented for representative rats in (A) the Miss, (B) Unilateral and (C) Bilateral groups. Mean optical densities are plotted for each group's hippocampus and overlying cortex at the level of the dorsal hippocampus. Note: error bars represent SEM. White scale bar, 2mm.

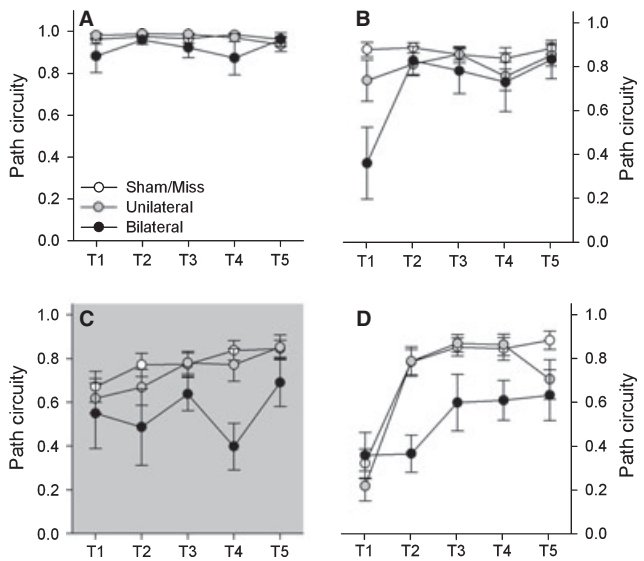

FIG. 4. Mean homeward segment path circuity is plotted for each group's five trials with (A) the cued refuge, (B) hidden refuge probe, (C) dark refuge probe, and (D) reversal probe. Note: Error bars represent SEM.

on path circuity failed to reveal a significant effect of Group ($F_{2,33} = 0.3$, $P = 0.743$), Trip ($F_{4,132} = 1.6$, $P = 0.178$) or Group × Trip interaction ($F_{8,132} = 0.4$, $P = 0.919$). The ANOVA conducted on time elapsed failed to reveal a significant effect of Group ($F_{2,33} = 1.0$, $P = 0.379$), Trip ($F_{4,132} = 0.7$, $P = 0.593$), and Group × Trip interaction ($F_{8,132} = 0.4$, $P = 0.919$). Groups did not significantly differ in the organization of their outward segments.

Topographical characteristics of the homeward segment were observed to differ among groups (see panel B of Fig. 4). The ANOVA conducted on path circuity revealed a significant effect of Group ($F_{2,33} = 6.6$, $P = 0.004$), Trip ($F_{4,132} = 5.6$, $P < 0.001$), and Group × Trip interaction ($F_{8,132} = 2.6$, $P = 0.011$). The nature of these differences was further examined by running a series of one-way ANOVAs on each trip. The ANOVA conducted on the first trip revealed a significant effect of Group ($F_{2,33} = 9.2$, $P = 0.001$), whereas no group differences were observed on subsequent trips. *Post hoc* analyses revealed that the Bilateral group had significantly more circuitous homeward segments on the first trip than did the Sham/Miss and Unilateral groups ($P < 0.05$). The disruption in the topography of the homeward segment associated with bilateral MTT lesion was restricted to the first trip.

Circular statistics further characterized group differences observed on the homeward segment during the hidden probe. The ANOVA conducted on mean vector length failed to reveal a significant effect of Group ($F_{2,33} = 0.4$, $P = 0.647$). This result permitted the use of circular statistics to examine group differences in heading direction. The Watson–Williams *F*-test failed to reveal a significant effect of Group ($F_{2,33} = 1.5$, $P = 0.238$) on heading direction. All groups' heading directions were clustered around the refuge (see panel B of Fig. 5).

Dark probe

During the dark probe, all rats carried the food pellet to the refuge for consumption. The topographic and temporal characteristics of the outward segment did not vary among groups. The ANOVA conducted on path circuity failed to reveal a significant effect of Group ($F_{2,33} = 0.4$, $P = 0.674$), Trip ($F_{4,132} = 0.4$, $P = 0.808$) or Group × Trip interaction ($F_{8,132} = 0.7$, $P = 0.691$). The ANOVA conducted on time elapsed failed to reveal a significant effect of Group ($F_{2,33} = 0.9$, $P = 0.416$), Trip ($F_{4,132} = 0.1$, $P = 0.982$) or Group × Trip interaction ($F_{8,132} = 1.0$, $P = 0.439$). Groups did not significantly differ in the organization of their outward segments.

Topographic characteristic of the homeward segment were observed to differ among groups (see panel C Fig. 4). The ANOVA conducted on path circuity revealed a significant effect of Group ($F_{2,33} = 5.2$, $P = 0.011$) and Trip ($F_{4,132} = 3.0$, $P = 0.021$); however, the Group × Trip interaction ($F_{8,132} = 0.9$, $P = 0.519$) was not significant. *Post hoc* analyses revealed that the Bilateral group had significantly more circuitous homeward segments than did the Sham/Miss or Unilateral groups ($P < 0.05$). In addition, a significant linear trend ($F_{1,33} = 7.3$, $P = 0.011$) was observed in path circuity across trips consistent with groups' homeward segments becoming more direct across trips.

Circular statistics were used to further characterize group differences observed on the homeward segment during the dark probe. The ANOVA conducted on mean vector length revealed a significant effect of Group ($F_{2,33} = 6.8$, $P = 0.003$). *Post hoc* analysis revealed that the Bilateral group was significantly more variable in heading direction than either the Sham/Miss or Unilateral groups ($P < 0.05$). Although all groups' average heading directions were orientated towards the refuge (see panel C of Fig. 5), significant group differences in mean vector length precluded further analysis of heading direction.

© 2010 The Authors. European Journal of Neuroscience © 2010 Federation of European Neuroscience Societies and Blackwell Publishing Ltd

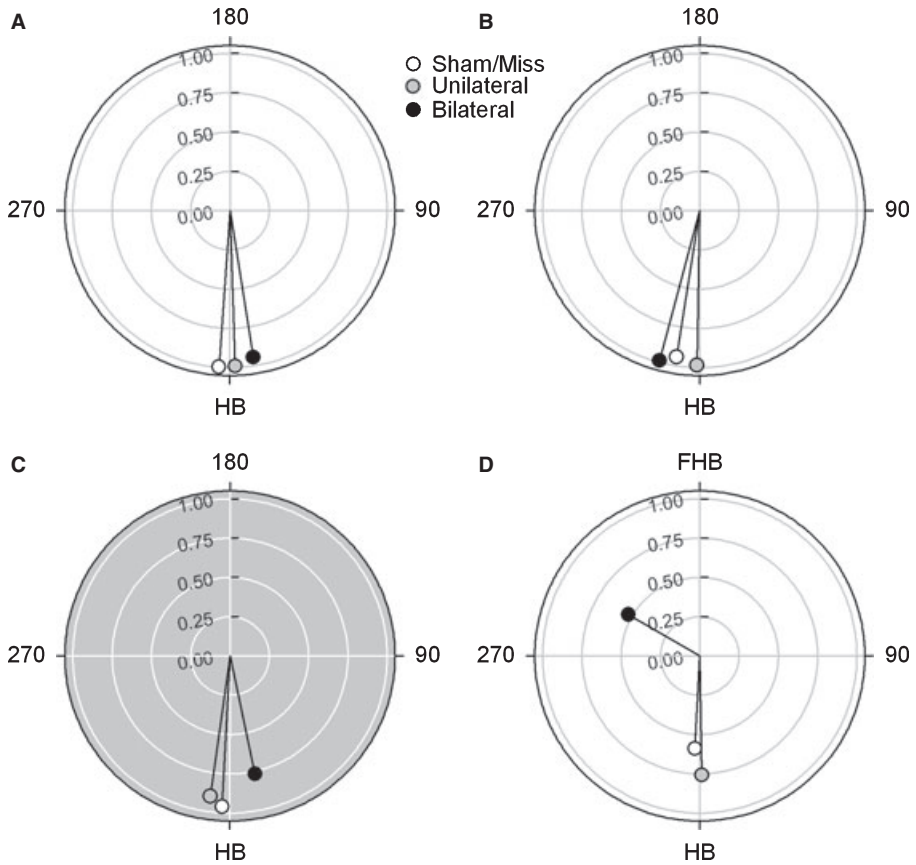

FIG. 5. Two characteristics of heading direction are plotted for the homeward segments with (A) the cued refuge, (B) hidden refuge probe, (C) dark probe and (D) reversal probe. The mean vector length or the average clustering of heading directions is plotted at the length of each ray. Average heading direction is plotted as the direction of each ray.

Reversal probe

During the reversal probe, all rats carried the food pellet to the refuge for consumption. The topographic and temporal characteristics of the outward segment were observed to vary among groups. The ANOVA conducted on path circuity revealed a significant effect of Group ($F_{2,33} = 4.1$, $P = 0.026$) and Trip ($F_{4,132} = 3.5$, $P = 0.009$); however, the Group × Trip interaction ($F_{8,132} = 1.4$, $P = 0.202$) was not significant. *Post hoc* analysis revealed that outward segments of the Unilateral group were significantly more circuitous than those observed in either the Sham/Miss or Bilateral groups (LSD, $P < 0.05$). In addition, outward segments became progressively less circuitous as revealed by a significant linear trend in path circuity across trips ($F_{1,33} = 7.1$, $P = 0.012$). A similar pattern of results was observed with time elapsed. The ANOVA conducted on time elapsed revealed a significant effect of Group ($F_{2,33} = 3.9$, $P = 0.030$) and Trip ($F_{4,132} = 5.1$, $P = 0.001$); however, the Group × Trip interaction ($F_{8,132} = 1.8$, $P = 0.082$) was not significant. *Post hoc* analysis revealed the outward segments of the Unilateral group were significantly longer in duration than either the Sham/Miss or Bilateral groups (LSD, $P < 0.05$). In addition, outward segments became progressively shorter in duration as revealed by a significant linear trend in time elapsed across trips ($F_{1,33} = 14.5$, $P = 0.001$).

Topographic characteristics of the homeward segment were observed to differ among groups (see panel D of Fig. 4). The ANOVA conducted on path circuity revealed a significant effect of Group ($F_{2,33} = 6.0$, $P = 0.006$), Trip ($F_{4,132} = 24.3$, $P < 0.001$) and Group × Trip interaction ($F_{8,132} = 2.2$, $P = 0.031$). *Post hoc* analyses revealed that the Bilateral group had significantly more circuitous homeward segments than did either the Sham/Miss or Unilateral groups (LSD, $P < 0.05$). In addition, homeward segments became progressively more direct as revealed by a significant linear trend in path circuity across trips ($F_{1,33} = 40.8$, $P < 0.001$). To characterize group difference as a function of trip, one-way ANOVAs were conducted on each trip. Significant group differences in homeward segment path circuity were restricted to trip 2 ($F_{2,35} = 6.7$, $P = 0.003$) and trip 3 ($F_{2,35} = 4.3$, $P = 0.021$). *Post hoc* analyses revealed that the Bilateral group had significantly more circuitous homeward segments than did the Sham/Miss or Unilateral groups (LSD, $P < 0.05$).

Circular statistics were used to further characterize group performance on the homeward segment during the reversal probe. The ANOVA conducted on the mean vector length failed to reveal a significant effect of Group ($F_{2,33} = 1.3$, $P = 0.286$). This result permitted the use of circular statistics to examine group differences in average heading direction. The Watson–Williams *F*-test revealed a significant effect of Group ($F_{2,33} = 6.1$, $P = 0.006$). Subsequent analyses revealed that the Bilateral group's average heading direction was significantly different from those observed in the Sham/Miss ($F_{1,22} = 9.6$, $P = 0.005$) and Unilateral ($F_{1,15} = 13.2$, $P = 0.002$) groups. Sham/Miss and Unilateral groups' average heading directions did not differ from each other and were oriented towards the new refuge location, whereas the Bilateral group's average heading direction was oriented towards the former location of the refuge (see panel D of Fig. 5).

© 2010 The Authors. European Journal of Neuroscience © 2010 Federation of European Neuroscience Societies and Blackwell Publishing Ltd

Water maze

Place training

During place training, rats swam to a hidden platform whose position did not change across days. The latency to reach the hidden platform decreased across days (see left-hand panel of Fig. 6). The ANOVA conducted on latency to reach the hidden platform revealed a significant effect of Group ($F_{2,33} = 3.6$, $P = 0.039$) and Block ($F_{4,132} = 102.4$, $P < 0.001$); however, the Group × Block interaction ($F_{8,132} = 0.6$, $P = 0.777$) was not significant. *Post hoc* analysis revealed that the Bilateral group had significantly longer latencies to find the hidden platform than did the Sham/Miss group (LSD, $P < 0.05$) but not the Unilateral group. In addition, latency to find the hidden platform significantly decreased across trials as indicated by a significant linear trend ($F_{1,33} = 250.5$, $P < 0.001$).

Matching-to-place

During matching-to-place testing, rats were given two trials to locate the hidden platform that shifted position across days. Collapsing across days, performance of the rats improved from trial 1 to trial 2 (see right-hand panel of Fig. 6). The ANOVA conducted on latency to reach the hidden platform revealed a significant effect of trial ($F_{1,33} = 56.7$, $P < 0.001$); however, the Group effect ($F_{2,33} = 2.8$, $P = 0.075$) and Group × Block interaction ($F_{2,33} = 2.3$, $P = 0.116$) were not significant. These results are consistent with rats learning the new location of the platform each day of matching-to-place testing.

Discussion

The current study demonstrated that MTT transection selectively disrupted performance on spatial tasks. First, performance observed during the food hoarding probes was consistent with MTT lesions impairing self-movement cue processing while sparing environmental cue use. Next, although MTT lesions attenuated performance in place training in the water maze, no differences in performance were observed during matching-to-place testing. This pattern of results is consistent with intact mnemonic functions. Finally, MTT lesions were observed to influence hippocampal cholinergic function. These results add to a growing literature demonstrating a role for limbic system structures in processing self-movement cues (Wallace *et al.*, 2008).

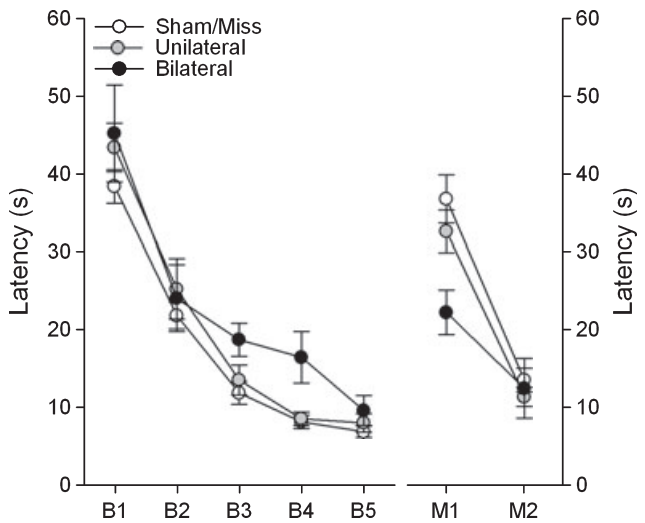

FIG. 6. Average latency to reach the hidden platform is plotted for the place training (left-hand panel) and matching-to-place testing (right-hand panel). Note: error bars represent SEM.

MTT transection selectively disrupts spatial orientation

Rats use environmental and self-movement cues to maintain spatial orientation. Debate has focused on the role of the limbic system in processing either source of information. One view has supported a role for the limbic system structures in encoding relationships between environmental cues (O'Keefe & Nadel, 1978; Morris *et al.*, 1982) and continues to guide research (Gupta *et al.*, 2009; Manns & Eichenbaum, 2009). In contrast, refined behavioral techniques that dissociate the use of different sources of information have been critical in demonstrating a role for the hippocampus (Whishaw, 1998), retrosplenial cortex (Whishaw *et al.*, 2001) and medial septum (Martin & Wallace, 2007) in processing self-movement cues. The results of the current study provide further support for the latter view, demonstrating a role for additional limbic system structures in processing self-movement cues.

During the food hoarding paradigm, rats carried food items to the refuge under conditions in which access to environmental cues were varied. During a probe session, performance on the homeward segment reflects the rats' ability to use either environmental or self-movement cues to guide movements. First, when presented with the cued refuge, all rats made direct returns to the refuge. This observation is consistent with rats using proximal cues associated with the refuge (i.e., beacon homing), distal cues associated with the testing room (i.e., piloting) or self-movement cues generated on the outward segment (i.e., dead reckoning) to return to the refuge. Next, all rats were able to directly return to the refuge when it was hidden below the surface of the table, consistent with rats using either distal room cues or self-movement cues to guide movement. One exception was the circuitous homeward segments of rats with bilateral MTT lesions observed on the first trial of the hidden probe. Disruption in performance on the first trial may reflect the Bilateral group's tendency to rely on proximal cues associated with the refuge to compensate for impaired self-movement cue processing. The direct returns observed on subsequent trials of the hidden probe are consistent with the sparing of the Bilateral group's ability to shift to using distal cues to guide navigation. Further, during the dark probe, rats were restricted to using self-movement cues (i.e., dead reckoning) to make direct returns to the refuge. Relative to the Sham/Miss and Unilateral groups, the Bilateral group exhibited significantly more circuitous homeward segments that were more variable in heading direction. This observation is consistent with MTT lesions impairing self-movement cue processing. Finally, environmental and self-movement cues can be placed in conflict by rotating the position of the hidden refuge to the opposite side of the table. No group differences were observed among groups on outward segment characteristics during the previous probes; however, the unilateral group took more time and more circuitous paths prior to finding the food pellet on the reversal probe than did the Sham/Miss and Bilateral groups. This result typically involved the Unilateral group visiting the former refuge location prior to finding the food pellet. Considering that neither the Sham/Miss nor Bilateral groups engaged in this behavior while searching for the food pellet, the conflicting environmental and self-movement cues might have produced a transient shift in navigation strategy used by the Unilateral group as they searched for the food pellet. After finding the food pellet on the first trial of the reversal probe, all rats typically visited the former refuge location, consistent with using distal room cues to guide movement. Subsequently, both Sham/Miss and Unilateral groups

© 2010 The Authors. European Journal of Neuroscience © 2010 Federation of European Neuroscience Societies and Blackwell Publishing Ltd

made direct returns to the new refuge location, consistent with using self-movement cues. In contrast, the Bilateral group returned to the former refuge location multiple times prior to returning to the new refuge location, and this tendency continued on subsequent trials. These observations provide further evidence that transection of the MTT selectively disrupt spatial orientation. The Bilateral group's perseveration to the former refuge location demonstrates spared use of environmental cues (i.e., piloting), whereas their failure to return to the new refuge location is consistent with impaired use of self-movement cues generated after exiting the refuge (i.e., dead reckoning). Performance during the food hoarding paradigm demonstrates that MTT lesions impair dead reckoning-based navigation while sparing the navigational strategies that depend on environmental cues.

Dead reckoning involves online processing of changes in angular and linear movements to estimate the direction and distance to the point where movement originated. Behavioral studies have demonstrated that direction and distance estimation are dissociable components (Etienne *et al.*, 1986; for review, see Wallace *et al.*, 2008). The results of the present study are consistent with bilateral MTT lesions disrupting self-movement cue processing, whereas unilateral MTT lesions spare the ability to use self-movement cues to return to the refuge. Dissociating whether MTT lesions impaired direction estimation, distance estimation or some combination of the two is not possible with the current set of behavioral tasks. However, previous studies employing recording and lesion techniques have reported that bilateral, but not unilateral, damage to the mammillary bodies is necessary to abolish the head direction signal in the ADN (Blair *et al.*, 1998, 1999). Taken together, it is possible that the mammillary bodies and their projections to the ADN are fundamental components of a direction estimation system associated with dead reckoning based navigation.

Role of self-movement cue processing in mnemonic function

In addition to spatial orientation, the MB have been implicated in mnemonic function (for reviews, see Vann & Aggleton, 2004; Vann, 2010). For example, damage that includes either the MB or the MTT is associated with anterograde amnesia in humans (Van der Werf *et al.*, 2003; Tsivilis *et al.*, 2008). Damage that includes these structures is often the result of degenerative (e.g., Korsakoff's syndrome) or acute (e.g., removal of colloid cysts) conditions that may result in hidden neuropathology. To further characterize the role of these structures in mnemonic function, researchers have used rats to examine the effects of more selective lesions on performance in spatial tasks. Damage to the MB or MTT has been observed to disrupt performance on the T-maze (Field *et al.*, 1978; Neave *et al.*, 1997; Vann & Aggleton, 2003), the water maze (Sutherland & Rodriguez, 1989; Vann & Aggleton, 2003) and the radial arm maze (Sziklas & Petrides, 1993; Sziklas & Petrides, 2000; Neave *et al.*, 1997; Vann & Aggleton, 2003). Disruptions in performance observed on these tasks have been attributed to impaired mnemonic function involving impoverished spatial encoding (Vann, 2010); however, several lines of evidence suggest that accurate self-movement cue processing may also contribute to mnemonic function (Whishaw & Wallace, 2003). For example, disorientating rats prior to training attenuates learning about relationships between environmental cues (Semenov & Bures, 1989; Biegler & Morris, 1996) and the formation of stable hippocampal place fields (Knierim *et al.*, 1995). Further, vestibular system damage in humans (Schautzer *et al.*, 2003; Brandt *et al.*, 2005) and rats (Russell *et al.*, 2003; Allen *et al.*, 2007; Zheng *et al.*, 2007) has been shown to disrupt performance on spatial memory tasks. Finally, brain structures associated with episodic memory (Vargha-Khadem *et al.*, 1997) are active during tasks that depend on self-movement cue processing (Wolbers *et al.*, 2007). Therefore, it is possible that the MB are involved in self-movement cue processing and this contributes to mnemonic function.

Water maze performance observed in the current study can also be interpreted as supporting a role for self-movement cue processing in mnemonic function. For example, during place training, MTT lesions attenuated learning the location of the hidden platform. Considering the effects of MTT lesions on performance in the food hoarding paradigm, the attenuated acquisition can be attributed to encoding relationships among environmental cues without the assistance of self-movement cue processing. In contrast, MTT lesions were not observed to significantly influence performance during matching-to-place testing. This result was unanticipated because self-movement cue processing would also be predicted to contribute to learning the new location of the hidden platform during matching-to-place testing. In addition, previous research has demonstrated that MTT lesions impair performance during matching-to-place testing (Vann & Aggleton, 2003). This disruption in performance was attributed to an impaired ability to encode new spatial information (Vann & Aggleton, 2003). Although a failure to encode new spatial information accounts for the attenuated acquisition associated with MTT lesions during place training, the failure to observe a disruption in performance during matching-to-place testing may depend on methodological differences between these studies. At least two factors may have contributed to the overall shorter latencies to find the hidden platform observed in the current study. First, rats were given two trials to find each hidden platform location in the current study and four trials to find each hidden platform location in the previous study (Vann & Aggleton, 2003). Giving more trials to search for the hidden platform increases the likelihood that rats will perseverate to the former platform location on subsequent days and increase the latency to reach the new platform location. Next, different rat strains were used in each study. Previous work has demonstrated that rats of the Long Evans strain have shorter latencies to locate the hidden platform during place training than rats of the Dark Agouti strain (Harker & Whishaw, 2002). Differences in performance are probably not due to varying levels of visual acuity between rat strains (Prusky *et al.*, 2002); however, no studies have yet characterized differences in self-movement cue processing among rat strains. The overall shorter latencies to find the hidden platform observed in the current study may have minimized the ability to detect group differences during matching-to-place testing. Therefore, dissociating whether MTT lesions impair encoding relationships among environmental cues in the water maze directly or via self-movement cue processing deficits remains open to debate.

MTT lesions alter neurobiology of the hippocampus

Several lines of evidence have implied a role for the mammillary bodies and their projections to the ADN in hippocampal function. First, in mice, mammillary body lesions have been shown to significantly reduce markers of cholinergic function in the hippocampus (Béracochéa *et al.*, 1995). Next, MTT lesions disrupt behaviorally induced c-Fos expression in the hippocampus (Vann & Albasser, 2009). Finally, in the present study, reductions in markers of cholinergic function were observed in the hippocampus; however, the overall reduction was less than that associated with immunotoxic lesions of the medial septum (Martin & Wallace, 2007). Both lesions have been shown to disrupt performance on tasks that depend on

© 2010 The Authors. European Journal of Neuroscience © 2010 Federation of European Neuroscience Societies and Blackwell Publishing Ltd

self-movement cue processing. Considering the potential that MTT lesions impair direction estimation, a double dissociation may be possible with selective cholinergic deafferentation and MTT lesions. Development of behavioral tasks that dissociate direction and distance estimation will significantly contribute to characterizing the role of these structures in spatial orientation.

Conclusion

This is the first study to examine the use of environmental and self-movement cues subsequent to MTT lesions. The performance impairments observed across the food hoarding and water maze tasks were consistent with MTT lesions impairing self-movement cue processing. These observations add to a growing literature demonstrating the necessity to examine the use of environmental and self-movement cues subsequent to manipulation of limbic system structures.

Acknowledgements

We would like to thank Patricia S. Wallace for comments on previous drafts of the manuscript. In addition, we would like to thank James V. Corwin for comments on the histology related to the project.

Abbreviations

AChE, Acetylcholinesterase; ADN, anterodorsal thalamic nucleus; DTN, dorsal tegmental nucleus of Gudden; MB, mammillary bodies; MTT, mammillothalamic tract; SUM, supramammillary nucleus.

References

Allen, K., Potvin, O., Thibaudeau, G., Doré, F.Y. & Goulet, S. (2007) Processing idiothetic cues to remember visited locations: hippocampal and vestibular contributions to radial-arm maze performance. *Hippocampus*, **17**, 642–653.

Barlow, J.S. (1964) Inertial navigation as a basis for animal navigation. *J. Theor. Biol.*, **6**, 76–117.

Bassett, J.P., Tullman, M.L. & Taube, J.S. (2007) Lesions of the tegmentomammillary circuit in the head direction system disrupt the head direction signal in the anterior thalamus. *J. Neurosci.*, **27**, 7564–7577.

Batschelet, E. (1981) *Circular Statistics in Biology.* Academic Press, London.

Béracochéa, D.J., Micheau, J. & Jaffard, R. (1995) Alteration of cortical and hippocampal cholinergic activities following lesion of the mammillary bodies in mice. *Brain Res.*, **670**, 53–58.

Biegler, R. & Morris, R. (1996) Landmark stability: studies exploring whether the perceived stability of the environment influences spatial representation. *J. Exp. Biol.*, **199**(Pt 1), 187–193.

Blair, H.T. & Sharp, P.E. (1996) Visual and vestibular influences on head-direction cells in the anterior thalamus of the rat. *Behav. Neurosci.*, **110**, 643–660.

Blair, H.T., Cho, J. & Sharp, P.E. (1998) Role of the lateral mammillary nucleus in the rat head direction circuit: a combined single unit recording and lesion study. *Neuron*, **21**, 1387–1397.

Blair, H.T., Cho, J. & Sharp, P.E. (1999) The anterior thalamic head-direction signal is abolished by bilateral but not unilateral lesions of the lateral mammillary nucleus. *J. Neurosci.*, **19**, 6673–6683.

Brandt, T., Schautzer, F., Hamilton, D.A., Brüning, R., Markowitsch, H.J., Kalla, R., Darlington, C., Smith, P. & Strupp, M. (2005) Vestibular loss causes hippocampal atrophy and impaired spatial memory in humans. *Brain*, **128**(Pt 11), 2732–2741.

Clark, B.J., Bassett, J.P., Wang, S.S. & Taube, J.S. (2010) Impaired head direction cell representation in the anterodorsal thalamus after lesions of the retrosplenial cortex. *J. Neurosci.*, **30**, 5289–5302.

Darwin, C. (1873) Origin of certain insects. *Nature*, **7**, 417–418.

Etienne, A.S. & Jeffery, K.J. (2004) Path integration in mammals. *Hippocampus*, **14**, 180–192.

Etienne, A.S., Maurer, R., Saucy, F. & Teroni, E. (1986) Short distance homing in the golden hamster after a passive outward journey. *Anim. Behav.*, **34**, 696–715.

Field, T.D., Rosenstock, J., King, E.C. & Greene, E. (1978) Behavioral role of the mammillary efferent system. *Brain Res. Bull.*, **3**, 451–456.

Frohardt, R.J., Bassett, J.P. & Taube, J.S. (2006) Path integration and lesions within the head direction cell circuit: comparison between the roles of the anterodorsal thalamus and dorsal tegmental nucleus. *Behav. Neurosci.*, **120**, 135–149.

Gallistel, C.R. (1990) *The Organization of Learning*. MIT, Cambridge.

Goodridge, J.P. & Taube, J.S. (1997) Interaction between the postsubiculum and anterior thalamus in the generation of head direction cell activity. *J. Neurosci.*, **17**, 9315–9330.

Goodridge, J.P., Dudchenko, P.A., Worboys, K.A., Golob, E.J. & Taube, J.S. (1998) Cue control and head direction cells. *Behav. Neurosci.*, **112**, 749–761.

Gupta, A.S., van der Meer, M.A., Touretzky, D.S. & Redish, A.D. (2009) Hippocampal replay is not a simple function of experience. *Neuron*, **65**, 695–705.

Hamilton, D.A., Rosenfelt, C.S. & Whishaw, I.Q. (2004) Sequential control of navigation by locale and taxon cues in the Morris water task. *Behav. Brain Res.*, **154**, 385–397.

Hamilton, D.A., Johnson, T.E., Redhead, E.S. & Verney, S.P. (2009) Control of rodent and human spatial navigation by room and apparatus cues. *Behav. Processes*, **81**, 154–169.

Harker, KT & Whishaw, IQ (2002) Place and matching-to-place spatial learning affected by rat inbreeding (Dark-Agouti, Fischer 344) and albinism (Wistar, Sprague-Dawley) but not domestication (wild rat vs Long-Evans, Fischer-Norway). *Behav. Brain Res.*, **134**, 467–477.

Kirk, I.J. (1997) Supramammillary neural discharge patterns and hippocampal EEG. *Brain Res. Bull.*, **42**, 23–26.

Kirk, I.J., Oddie, S.D., Konopacki, J. & Bland, B.H. (1996) Evidence for differential control of posterior hypothalamic, supramammillary, and medial mammillary theta-related cellular discharge by ascending and descending pathways. *J. Neurosci.*, **16**, 5547–5554.

Knierim, J.J., Kudrimoti, H.S. & McNaughton, B.L. (1995) Interactions between idiothetic cues and external landmarks in the control of place cells and head direction cells. *J. Neurophysiol.*, **80**, 425–446.

Maaswinkel, H., Jarrard, L.E. & Whishaw, I.Q. (1999) Hippocampectomized rats are impaired in homing by path integration. *Hippocampus*, **9**, 553–561.

Manns, J.R. & Eichenbaum, H. (2009) A cognitive map for object memory in the hippocampus. *Learn Mem.*, **16**, 616–624.

Martin, M.M. & Wallace, D.G. (2007) Selective hippocampal cholinergic deafferentation impairs self-movement cue use during a food hoarding task. *Behav. Brain Res.*, **183**, 78–86.

Means, L.W., Alexander, S.R. & O' Neal, M.F. (1992) Those cheating rats: male and female rats use odor trails in a water-escape "working memory" task. *Behav. Neural Biol.*, **58**, 144–151.

Morris, R.G., Garrud, P., Rawlins, J.N. & O'Keefe, J. (1982) Place navigation impaired in rats with hippocampal lesions. *Nature*, **297**, 681–683.

Murphy, J.J. (1873) Instinct: a mechanical analogy. *Nature*, **7**, 483.

Neave, N., Nagle, S. & Aggleton, J.P. (1997) Evidence for the involvement of the mammillary bodies and cingulum bundle in allocentric spatial processing by rats. *Eur. J. Neurosci.*, **9**, 941–955.

Neitz, J. & Jacobs, G.H. (1986) Reexamination of spectral mechanisms in the rat (*Rattus norvegicus*). *J. Comp. Psychol.*, **100**, 21–29.

O'Keefe, J. & Nadel, L. (1978) *The Hippocampus as a Cognitive Map*. Clarendon, Oxford.

Pearce, J.M., Roberts, A.D. & Good, M. (1998) Hippocampal lesions disrupt navigation based on cognitive maps but not heading vectors. *Nature*, **396**, 75–77.

Prusky, G.T., Harker, K.T., Douglas, R.M. & Whishaw, I.Q. (2002) Variation in visual acuity within pigmented, and between pigmented and albino rat strains. *Behav. Brain Res.*, **136**, 339–348.

Russell, N.A., Horii, A., Smith, P.F., Darlington, C.L. & Bilkey, D.K. (2003) Bilateral peripheral vestibular lesions produce long-term changes in spatial learning in the rat. *J. Vestib. Res.*, **13**, 9–16.

Schautzer, F., Hamilton, D., Kalla, R., Strupp, M. & Brandt, T. (2003) Spatial memory deficits in patients with chronic bilateral vestibular failure. *Ann. N Y Acad. Sci.*, **1004**, 316–324.

Semenov, L.V. & Bures, J. (1989) Vestibular stimulation disrupts acquisition of place navigation in the Morris water tank task. *Behav. Neural Biol.*, **51**, 346–363.

Stackman, R.W. & Taube, J.S. (1997) Firing properties of head direction cells in the rat anterior thalamic nucleus: dependence on vestibular input. *J. Neurosci.*, **17**, 4349–4358.

© 2010 The Authors. European Journal of Neuroscience © 2010 Federation of European Neuroscience Societies and Blackwell Publishing Ltd

Sutherland, R.J. & Rodriguez, A.J. (1989) The role of the fornix/fimbria and some related subcortical structures in place learning and memory. *Behav. Brain Res.*, **32**, 265–277.
Sziklas, V. & Petrides, M. (1993) Memory impairments following lesions to the mammillary region of the rat. *Eur. J. Neurosci.*, **5**, 525–540.
Sziklas, V. & Petrides, M. (2000) Selectivity of the spatial learning deficit after lesions of the mammillary region in rats. *Hippocampus*, **10**, 325–328.
Taube, J.S., Muller, R.U. & Ranck, J.B. (1990b) Head-direction cells recorded from the postsubiculum in freely moving rats II. Effects of environmental manipulations. *J. Neurosci.*, **10**, 436–447.
Tsivilis, D., Vann, S.D., Denby, C., Roberts, N., Mayes, A.R., Montaldi, D. & Aggleton, J.P. (2008) A disproportionate role for the fornix and mammillary bodies in recall versus recognition memory. *Nat. Neurosci.*, **11**, 834–842.
Van der Werf, Y.D., Scheltens, P., Lindeboom, J., Witter, M.P., Uylings, H.B. & Jolles, J. (2003) Deficits of memory, executive functioning and attention following infarction in the thalamus; a study of 22 cases with localised lesions. *Neuropsychologia*, **41**, 1330–1344.
Vann, S.D. (2005) Transient spatial deficit associated with bilateral lesions of the lateral mammillary nuclei. *Eur. J. Neurosci.*, **21**, 820–824.
Vann, S.D. (2010) Re-evaluating the role of the mammillary bodies in memory. *Neuropsychologia*, **48**, 2316–2327.
Vann, S.D. & Aggleton, J.P. (2003) Evidence of a spatial encoding deficit in rats with lesions of the mammillary bodies or mammillothalamic tract. *J. Neurosci.*, **23**, 3506–3514.
Vann, S.D. & Aggleton, J.P. (2004) The mammillary bodies: two memory systems in one? *Nat. Rev. Neurosci.*, **5**, 35–44.
Vann, S.D. & Albasser, M.M. (2009) Hippocampal, retrosplenial, and prefrontal hypoactivity in a model of diencephalic amnesia: evidence towards an interdependent subcortical-cortical memory network. *Hippocampus*, **19**, 1090–1102.
Vargha-Khadem, F., Gadian, G.D., Watkins, K.E., Connelly, A., Van Paesschen, W. & Mishkin, M. (1997) Differential effects of early hippocampal pathology on episodic and semantic memory. *Science*, **277**, 376–380.
Wallace, D.G., Gorny, B. & Whishaw, I.Q. (2002a) Rats can track odors, other rats, and themselves: implications for the study of spatial behavior. *Behav. Brain Res.*, **131**, 185–192.
Wallace, D.G., Hines, D.J., Pellis, S.M. & Whishaw, I.Q. (2002b) Vestibular information is required for dead reckoning in the rat. *J. Neurosci.*, **22**, 10009–10017.
Wallace, D.G., Martin, M.M. & Winter, S.S. (2008) Fractionating dead reckoning: role of the compass, odometer, logbook, and home base establishment in spatial orientation. *Naturwissenschaften*, **95**, 1011–1026.
Whishaw, I.Q. (1985) Formation of a place learning-set by the rat: a new paradigm for neurobehavioral studies. *Physiol. Behav.*, **35**, 139–143.
Whishaw, I.Q. (1998) Place learning in hippocampal rats and the path integration hypothesis. *Neurosci. Biobehav. Rev.*, **22**, 209–220.
Whishaw, I.Q. & Tomie, J. (1997) Piloting and dead reckoning dissociated by fimbria-fornix lesions in a rat food carrying task. *Behav. Brain Res.*, **89**, 87–97.
Whishaw, I.Q. & Wallace, D.G. (2003) On the origins of autobiographical memory. *Behav. Brain Res.*, **138**, 113–119.
Whishaw, I.Q., Maaswinkel, H., Gonzalez, C.L. & Kolb, B. (2001) Deficits in allothetic and idiothetic spatial behavior in rats with posterior cingulate cortex lesions. *Behav. Brain Res.*, **118**, 67–76.
Wolbers, T., Wiener, J.M. & Mallot HA, B.C. (2007) Differential recruitment of the hippocampus, medial prefrontal cortex, and the human motion complex during path integration in humans. *J. Neurosci.*, **27**, 9408–9416.
Zheng, Y., Goddard, M., Darlington, C.L. & Smith, P.F. (2007) Bilateral vestibular deafferentation impairs performance in a spatial forced alternation task in rats. *Hippocampus*, **17**, 253–256.

© 2010 The Authors. European Journal of Neuroscience © 2010 Federation of European Neuroscience Societies and Blackwell Publishing Ltd